

## EFFECT OF AQUEOUS EXTRACT OF *Mangifera indica* ON CARBON TETRACHLORIDE INDUCED LIVER DAMAGE OF WISTAR ALBINO RATS

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### ABSTRACT

The effect of aqueous extract of mango leaves (*Mangifera indica*) on carbon tetrachloride induced liver damage in wistar albino rats invitro was investigated. The liver function markers: Alanine transaminase (ALT), aspartate transaminase (AST); total protein (TP) and cholesterol were measured using standard protocols. The results showed that after the administration of carbon tetrachloride, the concentrations of ALT and AST in the serum increased with a spontaneous decrease in the total protein and cholesterol as a result of the damaged liver but on administration of varying concentrations of the extract for a period of seven days, there was a significant reduction in the concentrations of AST and ALT in the serum, while the total protein and cholesterol levels increased. The result indicated that the leaves of *M. indica* have hepatoprotective effect on carbon tetrachloride induced liver damage.

**Key words:** quinine-imine, biuret reagent, spontaneous decrease.

### INTRODUCTION

The use of herbal product for medical benefits has played an important role in nearly every culture on earth. Herbal medicine was practiced by ancient people of Africa, Asia, Europe and Americans (Djukanovic and Mach, 2001).

Carbon tetrachloride is one of the most potent hepatotoxins (toxic to the liver) and is widely used in scientific research to evaluate hepatoprotective agents (Sipes *et al.*, 2007). Trichloromethyl peroxide initiates the chain reaction of lipid peroxidation in the liver. Carbon tetrachloride intoxication leads to hypomethylation of cellular component which inhibits protein synthesis in the case of RNA (Schimizu *et al.*, 2003). This study

was therefore undertaken to evaluate the effect of *Mangifera indica* leaf extract on carbon tetrachloride (CCL<sub>4</sub>) induced liver lesion with a view to validating its therapeutic use in Folkloric medicine.

### MATERIALS AND METHODS

#### Collection of Plant and Preparation of Plant Extract

*M. indica* was obtained from a garden in Igbogo Road, Choba, Rivers State, Nigeria. The leaves were carefully plucked, thoroughly washed and were allowed to dry at room temperature until a constant weight was obtained. The dried sample was grounded with a blender and 100 g was transferred into an extraction bottle and 1000 ml of water added to the grounded

dried sample. After vigorous shaking for 5 minutes, the mixture was allowed to stand for 24 hours. The mixture was then filtered thrice, each time through a piece of white clean cotton cloth. A final filtration using Whatman No. 541 filter paper followed. The filtrate was then stored in the refrigerator and then 20 and 25 % dilutions were prepared from the stock.

### **Experimental Animals**

A total of sixteen (16) wistar albino rats (about 8 months old), weighing  $210 \pm 9.0$  g were obtained from the Department of Zoology and Environmental Biology, University of Nigeria, Nsukka, Enugu State, and transported to the Department of Biochemistry of Port Harcourt Animal House. They were housed in cages (4 rats per cage) in the animal house laboratory to acclimatize while they received their standard diet (normal feed) (Livestock Feeds, Nigeria) and water *ad libitum*. The standard guidelines for the use of experimental animals (including applying humane actions during sacrifice) were adhered to.

### **Treatment of Animals**

The animals were divided into four (4) experimental groups, each consisting of four (4) rats. The animals were allowed to acclimatize for a period of seven days at the end of which CCL<sub>4</sub> was administered to the rats in groups 2, 3 and 4. Liver damage was induced by the oral administration of CCL<sub>4</sub> diluted with vegetable oil (1:1) at 0.5 mg/kg body wt. except for the control animals (no liver damage) (Rao and Mishra, 1998). The CCL<sub>4</sub> was dissolved in vegetable oil in a 1.1 ratio and was administered to the animals orally at a volume of 2.5 ml/kg respectively.

Treatment with the plant's extract started after 48 hours post CCL<sub>4</sub> administration and lasted for seven days.

The treatment of different experimental groups is as follows:

**Group 1 (Normal Control):** The animals in this group were fed with rat pellets and water only.

**Group 2 (Positive Control):** Those in this group were administered with 2.5 ml/kg of CCL<sub>4</sub> + Vegetable oil (1: 1 V/V), their normal feed and water.

**Group 3 (Test 1):** The animals in this group were administered with 2.5 ml/kg CCL<sub>4</sub> + Vegetable oil and 2.0 ml/kg of plant extract, their normal feed and water.

**Group 4 (Test 2):** The animals in this group were administered with 2.5 ml/kg CCL<sub>4</sub> + Vegetable oil and 2.5 ml/kg of plant extract, their normal feed and water.

Rats were anaesthetized in chloroform and sacrificed twenty-four hours after the last treatment (for groups 3 and 4). The thoracic region was opened to expose the heart and fresh blood was collected by cardiac puncture. Serum obtained was analyzed in the Chemical Pathology Laboratory, University of Port Harcourt Teaching Hospital.

### **Enzyme Assays**

The determination of aspartate aminotransferase in the serum samples were performed at 37°C using the Randox kit by measuring the amount of oxaloacetate hydrazone formed in the presence of L-aspartate,  $\alpha$ -oxoglutarate and 2,4-dinitrophenyl hydrazine as reported by Ibekwe *et al.* (2007). For alanine

aminotransferase, L-alanine replaced L-aspartate and pyruvate replaced oxaloacetate. The determination of total protein involves the reaction of the peptide bonds of protein with biuret reagent in an alkaline medium to give a purple colour. The determination of the cholesterol was by enzymatic hydrolysis and oxidation method. The indicator quinine-imine is formed from hydrogen peroxide and 4-amino anti-pyrene in the presence of the phenol and peroxidase (Third Report of the National Cholesterol Education Programme (NCEP) Expert Panel, 2001).

### Statistical Analysis

All data were expressed as mean  $\pm$  SEM (Standard error of mean) and statistically analyzed with ANOVA (Analysis of variance) at 95 % confidence level. A p value of  $< 0.05$  was considered statistically significant.

### RESULTS

The results obtained for the serum activities (U/L) of AST, ALT, total protein and cholesterol in rats following CCL<sub>4</sub> induced damage of hepatocytes and administration of aqueous extract of leaves of *M. indica* are shown in Table 1.

**Table 1: Comparison of the controls and tests result for the liver function markers, total protein and cholesterol**

	AST	ALT	TP	Cholesterol
Normal control	47.00 $\pm$ 2.94	53.75 $\pm$ 2.02	75.50 $\pm$ 3.01	52.43 $\pm$ 2.18
Positive control	96.00 $\pm$ 1.83	92.25 $\pm$ 1.65	60.50 $\pm$ 2.22	61.43 $\pm$ 2.18
Test 1	78.00 $\pm$ 1.15	72.67 $\pm$ 1.76	68.33 $\pm$ 1.45	61.83 $\pm$ 1.03
Test 2	66.00 $\pm$ 2.08	64.67 $\pm$ 1.86	68.67 $\pm$ 2.03	61.90 $\pm$ 2.06

### DISCUSSION

The animals induced with CCL<sub>4</sub> had significant hepatic damage as elicited by the elevated levels of hepatic serum markers. The post treatment with aqueous extract of *M. indica* leaves significantly reduced the protein and cholesterol. This finding was in agreement with studies carried out by other researchers on hepatoprotective effects of plant extracts such as; *Solanum americanum* (Kumar *et al.*, 2011).

The overall experimental study indicate that aqueous extract of *M. indica* leaves helped in ameliorating the damaging action of carbon tetrachloride in the liver of rats.

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